Cholesteryl Ester Transfer Protein and Lecithin:Cholesterol Acyltransferase Activities in Hispanic and Anglo Postmenopausal Women: Associations With Total and Regional Body Fat

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Reverse cholesterol transport is one process by which high-density lipoprotein (HDL) cholesterol has been hypothesized to play a role in reducing the risk of coronary heart disease. This study was designed to examine cholesteryl ester transfer protein (CETP) and lecithin:cholesterol acyltransferase (LCAT) activities, 2 modulators of reverse cholesterol transport, in Hispanic and Anglo postmenopausal women. The associations between plasma CETP and LCAT activities and body composition were also examined. Of the 199 subjects, 33% were of Hispanic origin and 47% were undergoing hormone replacement therapy (HRT). Body composition was measured by dual-energy x-ray absorptiometry (DXA) and anthropometry. Plasma CETP activity was higher in Hispanic compared to Anglo women, although the difference was eliminated when data were adjusted for abdominal fat. Hispanic women had lower plasma HDL cholesterol concentrations, higher total cholesterol:HDL cholesterol ratios and triglyceride concentrations, and greater susceptibility of low-density lipoprotein (LDL) particles to oxidation. Hispanic women also had a significantly greater relative deposition of body fat on the trunk and intra-abdominally than did Anglo women, even after adjusting for total body fat. There were no significant ethnic differences in LCAT activity. Plasma CETP and LCAT activities were negatively correlated with HDL cholesterol and positively correlated with total cholesterol, LDL cholesterol, and triglycerides, as well as total and regional body composition variables. In conclusion, results suggest a greater risk for coronary heart disease in Hispanic women compared to Anglo women.

CORONARY HEART DISEASE is the leading cause of death in women, with the risk increasing after menopause.[1,2] Coronary heart disease risk factors in women include high plasma triglyceride and low high-density lipoprotein (HDL) cholesterol concentrations. HDL cholesterol is a modifiable risk factor that is inversely related to the prevalence and incidence of coronary heart disease,[3,4] as well as the severity of atherosclerosis.[5] The protection offered by HDL cholesterol against the development of coronary heart disease is hypothesized to be due to the role HDL plays in reverse cholesterol transport.[6] Lecithin:cholesterol acyltransferase (LCAT) and cholesteryl ester transfer protein (CETP) are intravascular factors involved in reverse cholesterol transport.[7,8] LCAT is associated with HDL and esterifies cholesterol in HDL particles. CETP facilitates the exchange of cholesterol esters from HDL with triglyceride from apoprotein B–containing lipoproteins. An increase in CETP activity may be atherogenic in hyperlipidemic individuals because of the potential for the deposition of cholesteryl ester from apoprotein B–containing lipoproteins (circulating in excess) in arteries, promoting lesion development.[9]

Cardiovascular mortality rates are higher in Hispanic women than in Anglo women.[2] This difference in the incidence of disease may be due to differences in the presence or severity of coronary heart disease risk factors. Ethnic differences in serum lipids and lipoproteins have been reported in women independent of age, body mass index, alcohol intake, smoking, exercise, hormone replacement therapy (HRT), and oral contraceptive use.[10-12] For example, HDL cholesterol concentrations have previously been found to be lower in Hispanic compared to Anglo women.[10-12] Ethnic differences in body fat distribution, as well as the association between body fat distribution and lipids and lipoproteins, have also been reported in women. For example, Mexican American women deposit a greater proportion of body fat on their trunk, while Anglo women preferentially deposit fat in the gluteal-femoral area.[12,13]

Sex steroids have been found to alter body composition and thus may influence coronary heart disease risk.[14] Premenopausal women tend to deposit fat in the gluteal-femoral area, which may confer a protection against coronary heart disease risk.[15] However, the relative proportion of intra-abdominal adipose tissue in women increases with age, particularly after menopause.[14,16] Intra-abdominal deposition of fat is associated with lower concentrations of HDL cholesterol and increased risk of coronary heart disease.[15,17] HRT may offer protection against the postmenopausal increase in abdominal fat.[18,19] Plasma LCAT and CETP activities have not been investigated with regard to body fat deposition. As HDL cholesterol levels are influenced by body composition, and LCAT and CETP activities modulate HDL cholesterol concentrations, the activities of these factors may be influenced by body fat distribution.

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Submitted January 8, 2002; accepted September 24, 2002.

Supported in part by a grant from the Arizona Disease Control Research Commission, Phoenix, AZ (9511) and by NIH Grant No. AR39939. Part of the research upon which this publication is based was performed pursuant to Contract No. N01-WH-3-201115 with the NIH, DHHS.

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0026-0495/03/5203-0003$30.00/0

doi:10.1053/meta.2003.50045

The objectives of this cross-sectional study were (1) to examine differences in plasma CETP and LCAT activities in Hispanic and Anglo women, and (2) to examine the associations between CETP and LCAT activities and body composition in these 2 ethnicities.

MATERIALS AND METHODS

Subjects

This study was designed as an ancillary study to the Women's Health Initiative (WHI) and the Bone Estrogen Strength Training (BEST) Study being conducted at the Arizona Prevention Center and the Body Composition Laboratory, Department of Physiology, respectively, at the University of Arizona in Tucson, AZ. Only participants assigned to the Observational Arm of the WHI were asked to participate in the present study. Informed consent was obtained from all subjects before participation in this study. The study was approved by the University of Arizona Institutional Review Board for research on human subjects. The initial clinic visit for this study coincided with the first screening visit required by the parent studies. Women were excluded from participation if they had been diagnosed with cardiovascular disease or were taking lipid-lowering agents, diagnosed with thyroid dysfunction or disease-related causes for obesity, diagnosed with insulin-dependent diabetes mellitus, or had a body mass index (BMI) less than the 5th or greater than the 95th age-related percentile (Hispanic Health and Nutrition Examination Survey [HHANES] and National Health and Nutrition Examination Survey [NHANES]) from measured height and weight.[20,21] Subjects were included in the study if they had not used HRT in the last year, or if they were using estrogen only or estrogen plus a progestin for at least 3 months. Women between 40 and 79 years of age were included in the study. Sixty-six (33%) women were Hispanic and 133 (67%) were non-Hispanic white.

Menopausal status and duration of menopause were self-reported by participants in the BEST study and the WHI. Women recruited for the BEST study were between the ages of 40 and 65 years and between 3 and 10.9 years postmenopausal (time since last menstrual bleeding). A menopausal algorithm was used to ascertain eligibility for WHI participants (Fig 1). Participants were asked to bring in all medication bottles, thus if a woman reported using HRT, the type and amount could be ascertained. Fifty-three percent of the non-Hispanic white and 36% of the Hispanic participants reported using HRT. Compliance to HRT was not determined for either study.

Body Composition

Body composition was assessed by whole body dual-energy x-ray absorptiometry (DXA) using the Hologic QDR-2000 Bone Densitometer (Hologic, Bedford, MA; Version 5.60A) or the Lunar model DPX-L (Lunar, Madison, WI; Version 1.3Y). Two scans were preformed on medium speed using the Lunar DXA and the mean of those scans was used in analyses. One-array whole body scan was performed using the Hologic DXA. Radiation exposure for a total body scan was 0.03 mrem and 1.5 mrem for Lunar and Hologic, respectively. Abdominal adipose tissue deposition was determined using software provided by Hologic (Version 7.10C) or Lunar (Version 1.3Y, Extended Research mode). An abdominal region of interest comprising all abdominal tissue was analyzed between, and inclusive of, the first and fourth lumbar intervertebral disks.

Body circumferences and skinfolds were measured following standardized procedures.[22] Skinfolds were measured at the triceps, suprailiac, subscapular, abdomen, and thigh. Circumferences were measured at the waist, abdomen, buttocks, and thigh. Three measurements were taken at each site and the mean of the 3 trials was used in data analysis. Intra-abdominal adipose tissue was estimated using the equations of Svendsen et al,[23] which includes measured abdominal adipose tissue deposition from whole body DXA scans, trunk skinfold measurements (suprailiac, subscapular, and abdomen), and waist-to-hip ratio (WHR). Weight and height were measured twice in light clothing without shoes and recorded to the nearest 0.1 kg and 0.1 cm, respectively.

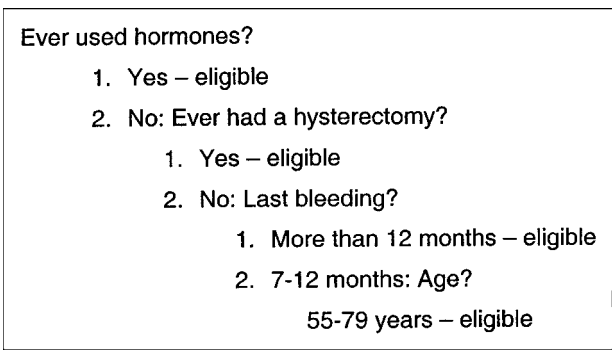

Fig 1. Women's Health Initiative menopausal algorithm.

Lipids and Lipoproteins

Fasting blood samples were collected twice within a 6-week period, to minimize intra-individual variability.[24,25] After a 12-hour overnight fast, 40 mL of blood was collected via venipuncture using vacutainer tubes containing EDTA. Blood was immediately put on ice until centrifugation at 2,600 rpm for 30 minutes. Plasma was removed from vacutainer tubes and a mixture of preservatives was added (sodium azide [0.1 mg/mL], aprotonin [0.5 mg/mL], and phenylmethanesulfonyl flouride [PMSF; 0.1 mg/mL]).

Total plasma cholesterol and HDL cholesterol concentrations were quantified by an enzymatic method[26] using a High Performance Cholesterol reagent (Boehringer Mannheim Diagnostics, Indianapolis, IN). HDL cholesterol was first isolated using the $MgCl_2$-dextran sulfate method for the precipitation of apoprotein B lipoproteins.[27] Free cholesterol was determined by an enzymatic method using a kit from Wako Pure Chemical Industries (Richmond, VA). Triglyceride concentrations were quantified by an enzymatic method using a Boehringer Mannheim Diagnostics commercial test kit that adjusts for free glycerol.[28] Low-density lipoprotein (LDL) cholesterol was calculated using the Friedwald equation.[29]

The susceptibility of LDL particles to oxidation was determined on a subset of subjects. Copper-mediated oxidation of LDL was performed by adding a 0.5 mmol/L $CuCl_2 \cdot 2H_2O$ solution to 0.2 mg protein/mL LDL. The extent of oxidation was measured by incubating samples for 3 hours at 37°C. The lipid peroxide content of oxidized LDL was determined by the analysis of thiobarbituric acid reactive substances (TBARS) expressed as malonaldehyde (MDA) equivalents.[30] The TBARS assay was conducted by adding 2 mL of TBARS reagent (26 mmol/L TBA, 0.92 trichloracetic acid in 0.25N HCl) to a 550-μL incubation mixture and heating for 15 minutes. Tubes were removed from the water bath, 0.025 L of n-butanol was added, tubes were shaken, and phases were separated by centrifugation at 1,500 $\times$ g for 15 minutes. A pink color developed in the organic layer and samples were read in a spectrophotometer at 532 nm.

CETP and LCAT Activities

Physiological CETP activity was determined by analysis of the decrease in HDL cholesteryl ester mass, without inhibiting the de novo synthesis of cholesterol esters by LCAT. CETP activity was determined on fresh plasma for analysis of the mass transfer of cholesteryl ester between HDL and apoprotein B–containing lipoproteins.[31] Samples

were incubated at 37°C for 0, 3, and 6 hours in a water bath. HDL cholesterol was isolated and samples were frozen at −20°C until plasma and HDL total and free cholesterol concentrations were determined in triplicate for 0, 3, and 6 hours as described above. Plasma LCAT activity was determined as the change in plasma free cholesterol between 0, 3, and 6 hours at 37°C.[32] Samples were frozen at −20°C until plasma free cholesterol was determined in triplicate for the 0-, 3-, and 6-hour samples as described above. The slopes of the lines over the 6-hour period were used to determine CETP and LCAT activities.

Plasma lipid and lipoprotein concentrations, and LCAT and CETP activities were measured in the Lipid Metabolism Laboratory at the University of Arizona. Standardization and quality control for plasma and lipoprotein cholesterol and triglyceride assays were maintained by participation in the Centers for Disease Control-National Heart, Lung and Blood Institute (CDC-NHLBI) Lipid Standardization Program. Certification was renewed quarterly by the use of blinded samples and has been maintained in the Lipid Metabolism Laboratory since 1989.

Statistical Methods

A total of 199 women were included in this study, 122 from the BEST study and 77 from the WHI. Two blood draws were obtained on 176 of the 199 women. When 2 blood draws were available the mean was used in analyses, otherwise results from a single draw was used. Because of technical difficulties in the collection of blood samples or the performance of laboratory assays, not every plasma variable is based on a sample size of 199 (range, 193 to 199). The susceptibility of LDL particles to oxidation was determined on a subsample of 78 subjects. Total and regional body composition measures were obtained on 186 to 199 of the subjects due to scheduling difficulties.

Data were analyzed using the BMDP statistical software (Version 7.0; SPSS, Chicago, IL). Pearson correlation coefficients were used to estimate relationships between plasma CETP and LCAT activities and total and regional body composition variables, as well as plasma lipid and lipoprotein variables. Analysis of covariance (ANCOVA) was used to determine differences in plasma CETP and LCAT activities between ethnic groups, using years of HRT usage, age, and body composition variables as covariates. Data are presented as the mean ± SD. Data were log-transformed if there was a significant difference in the variance between the 2 groups.

RESULTS

Correlations, standard errors of estimate (SEE), and *t* tests were calculated to compare day 1 and day 2 abdominal fat mass (kg and %) using the Lunar DXA in a subsample of 109 subjects.[33] Between-day correlations for fat mass (kg and %) were 0.992 and 0.976, respectively. SEEs for fat mass (kg and %) were 0.17 and 1.99, respectively. Small, significant mean differences ($P < .05$) found between days for fat mass (kg and %) were −0.39% and −0.045 kg, respectively. These differences were due to the high precision of DXA and are not physiologically relevant. A comparison of total body fat (kg and %) was done between the Lunar and Hologic absorptiometers in 20 women between the ages of 25 and 62 years.[34] The correlation coefficient between the 2 absorptiometers was 0.99 for both total body percent fat and total body fat (kg). Total body fat (kg) differences between the 2 absorptiometers was 0.37%.

Lipids and Lipoproteins

Table 1 contains descriptive data for plasma variables. Significant ethnic differences were found with Hispanic women having higher mean plasma CETP activity, total cholesterol: HDL cholesterol ratio and triglycerides, and a lower mean HDL cholesterol concentration than Anglo women. The oxidative susceptibility of LDL particles was also significantly greater in Hispanic women. Additionally, Hispanic women were significantly older than Anglo women.

Table 1. Plasma Variables for Hispanic and Anglo Postmenopausal Women

	Hispanic Women (n = 66)	Anglo Women (n = 133)
Age (yr)	59.0 ± 7.8§	56.6 ± 6.5
Years postmenopausal	11.6 ± 10.7	8.7 ± 6.7
Years on HRT	3.3 ± 3.3	4.7 ± 6.1
CETP (μg/mL · h)	14.5 ± 8.3	11.4 ± 9.0
LCAT (μg/mL · h)	26.6 ± 6.4	25.6 ± 7.5
Total cholesterol (mg/dL)	223 ± 43	222 ± 37
HDL cholesterol (mg/dL)	52 ± 15‡	62 ± 14
LDL cholesterol (mg/dL)	138 ± 34 (64)	135 ± 33 (123)
Total:HDL cholesterol ratio	4.6 ± 1.4‡	3.8 ± 1.1
Triglycerides (mg/dL)	168 ± 100† (64)	129 ± 86 (127)
LDL oxidation (MDA; nmol/mg LDL protein)	40.7 ± 16.6† (49)	30.3 ± 10.9 (29)

NOTE. Values are mean ± SD, with sample size in parentheses.
Significant differences: *$P \leq .05$; †$P \leq .01$; ‡$P \leq .001$.
Log-transformed data P value: §$P \leq .05$.

Correlation coefficients were calculated to evaluate the relationships between plasma CETP and LCAT activities and plasma lipid and lipoprotein measures for the total sample and ethnic subsamples (Table 2). In the total sample, a significant negative correlation was found between CETP activity and HDL cholesterol and positive correlations were found between CETP activity and total cholesterol, LDL-C, total cholesterol: HDL cholesterol ratio, triglyceride concentrations, and the oxidative susceptibility of LDL particles (Table 2). Correlation coefficients tended to be similar in magnitude to the total sample in the Anglo and Hispanic subsample for total cholesterol, LDL-C, and triglycerides, although correlations for LDL-C were not significant for either ethnicity. The correlations between CETP activity and oxidative susceptibility of LDL particles were not significant in either ethnic subsample (Table 2).

Moderate correlation coefficients were observed between plasma LCAT activity and plasma lipids and lipoproteins. All plasma variables were significantly related to LCAT activity in the entire sample. Significant correlations were found in the Hispanic subsample for all variables except HDL cholesterol and oxidative susceptibility of LDL particles, while all variables except LDL cholesterol and oxidative susceptibility of LDL particles were significantly related to plasma LCAT activity in the Anglo subsample (Table 2).

Age was not significantly related to plasma CETP in the total sample or the Hispanic subsample, but did have a significant negative association with CETP activity in the Anglo subsample. Age was also found to have a significant negative relationship with plasma LCAT activity in the total sample and the Anglo subsample (Table 2). Years postmenopausal was not significantly related to either CETP ($r = 0.01$) or LCAT ($r = -0.11$) activities and years of HRT usage was not significantly related to either CETP ($r = -0.07$) or LCAT ($r = -0.09$)

Table 2. Correlations of CETP and LCAT With Plasma Variables in Total Sample, and in Hispanic and Anglo Subsamples

	CETP (μg/mL · h)			LCAT (μg/mL · h)		
	Total (N = 199)	Hispanic (n = 66)	Anglo (n = 133)	Total (N = 199)	Hispanic (n = 66)	Anglo (n = 133)
Age (yr)	−0.10	−0.02	−0.18*	−0.15*	−0.03	−0.23†
Total cholesterol (mg/dL)	0.22†	0.26*	0.21*	0.30‡	0.49‡§	0.21*§
HDL cholesterol (mg/dL)	−0.26‡	−0.39‡	−0.15	−0.19†	−0.16	−0.19*
LDL cholesterol (mg/dL)	0.15* (187)	0.20 (64)	0.12 (123)	0.20† (187)	0.33† (64)	0.14 (123)
Total:HDL cholesterol	0.40‡	0.55‡§	0.28‡§	0.36‡	0.45‡	0.32‡
Triglycerides (mg/dL)	0.41‡ (191)	0.42‡ (64)	0.38‡ (127)	0.39‡ (191)	0.50‡ (64)	0.32‡ (127)
LDL oxidation (MDA; nmol/mg LDL protein)	0.28* (78)	0.21 (49)	0.14 (29)	0.25* (78)	0.18 (49)	0.12 (29)

NOTE. Samples sizes are given in parentheses.

*$P \leq .05$; †$P \leq .01$; ‡$P \leq .001$.

§Significant test for ethnic differences between independent correlations ($P \leq .05$).

activities. Mean values for years postmenopausal and years receiving HRT in the entire sample were 9.4 ± 8.1 and 4.3 ± 5.5, respectively.

Tests for significant ethnic differences between independent correlations were performed using z scores. The correlations for total cholesterol:HDL cholesterol ratio and CETP activity and total cholesterol and LCAT activity in Hispanic vs. Anglo subsamples were significantly different ($P \leq .05$) from one another. No other correlation coefficients in the subsamples were significantly different ($P \leq .05$).

Body Composition

Descriptive data for total and regional body composition variables are reported in Table 3. Significant ethnic differences were observed for all regional and total body composition measures except for weight and total body fat mass (kg). Hispanic women had higher mean intra-abdominal adipose tissue area, relative (%) and absolute (kg) abdominal adipose tissue estimated by DXA, WHR, trunk:total fat ratio as measured by both DXA and skinfolds, and BMI compared to Anglo women. Hispanic women were also significantly shorter in stature than Anglo women.

Table 3. Total and Regional Body Composition Variables for Hispanic and Anglo Postmenopausal Women

	Hispanic Women (N = 66)	Anglo Women (N = 133)
Height (cm)	158.2 ± 5.6‡	163.1 ± 6.0
Weight (kg)	68.1 ± 11.9	68.4 ± 11.6
BMI (kg/m^2)	27.2 ± 4.4*	25.7 ± 4.1
Waist circumference (cm)	86.0 ± 11.0†	81.6 ± 10.4
WHR	0.84 ± 0.08‡	0.79 ± 0.07
Skinfold trunk:total ratio	0.60 ± 0.08‡	0.56 ± 0.08
Total body fat (kg)	28.3 ± 9.0 (57)	27.2 ± 8.7 (129)
Total body fat (%)	41.6 ± 7.4* (57)	39.4 ± 6.8 (129)
Abdominal fat (kg)	4.0 ± 1.8† (57)	3.1 ± 1.4 (126)
Abdominal fat (%)	41.3 ± 9.6† (57)	37.1 ± 9.5 (126)
IAAT (cm^2)	174.2 ± 84.6‡ (55)	125.1 ± 71.2 (126)
DXA trunk:total ratio	0.14 ± 0.04‡ (57)	0.11 ± 0.02 (126)

NOTE. Values are mean ± SD, with sample sizes given in parentheses.

Abbreviation: IAAT, intra-abdominal adipose tissue.

Significant differences: *$P \leq 0.05$; †$P \leq .01$; ‡$P \leq .001$.

The relationships between plasma CETP and LCAT activities and total and regional body composition measures were also examined (Table 4). Significant positive relationships were found between CETP activity and all body composition measures, with the exception of height, in the entire sample and in the Anglo subsample. Only regional body composition measures; intra-abdominal adipose tissue, trunk:total ratios (skinfold and DXA), abdominal fat (kg and %), WHR, waist circumference; were significantly related to CETP activity in the Hispanic subsample. Similar results were noted when the relationships among body composition measures and plasma LCAT activity were evaluated. The exceptions were that WHR and waist circumference were not significantly related to plasma LCAT activity and that total body fat percentage was related to LCAT activity in the Hispanic subsample. No significant ethnic differences were found for correlation coefficients among plasma CETP and LCAT activities and total and regional body composition measures using z scores (Table 4).

Multiple Regression Analysis

Stepwise multiple regression analyses were performed to determine which plasma and body composition variables best predicted plasma CETP and LCAT activities. All plasma and body composition variables, as well as ethnicity, HRT usage (yes/no), age, years on HRT, and years postmenopausal were included in the analyses. The results are presented in Table 5. The best predictors of CETP activity were ethnicity, plasma triglyceride concentration, and total cholesterol:HDL cholesterol ratio, accounting for 46% of the variance in CETP activity. The best predictors of LCAT activity were ethnicity, total cholesterol:HDL cholesterol ratio, BMI, and WHR, accounting for 45% of the variance in LCAT activity. In both CETP and LCAT activity models, ethnicity was the best predictor of activities. Body composition measures, BMI and WHR, were significant predictors in the model for LCAT activity only. The combination of predictor variables were logical predictors for CETP and LCAT activities and accounted for 46% and 45% of the variability, respectively. Other unmeasured factors, dietary or behavioral, may lead to a reduction in biological variability, and hence a further reduction in the SEE.

Table 4. Correlations of CETP and LCAT With Total and Regional Body Composition in Total Sample, and in Hispanic and Anglo Subsamples

	CETP (μg/mL · h)			LCAT (μg/mL · h)		
	Total (N = 199)	Hispanic (n = 66)	Anglo (n = 133)	Total (N = 199)	Hispanic (n = 66)	Anglo (n = 133)
Height (cm)	−0.07	0.01	−0.01	−0.07	−0.05	−0.04
Weight (kg)	0.30‡	0.23	0.34‡	0.27‡	0.11	0.34‡
BMI (kg/m^2)	0.34‡	0.24	0.37‡	0.31‡	0.14	0.38‡
Waist circumference (cm)	0.37‡	0.27*	0.39‡	0.40‡	0.18	0.48‡
WHR	0.30‡	0.25*	0.27†	0.38‡	0.20	0.46‡
Skinfold trunk:total ratio	0.38‡	0.25*	0.40‡	0.44‡	0.29*	0.49‡
Total body fat (kg)	0.28‡ (186)	0.22 (57)	0.30‡ (129)	0.30‡ (186)	0.24 (57)	0.32‡ (129)
Total body fat (%)	0.24‡ (186)	0.23 (57)	0.22* (129)	0.25‡ (186)	0.28* (57)	0.24† (129)
Abdominal fat (kg)	0.37‡ (183)	0.29* (57)	0.39‡ (126)	0.39‡ (183)	0.34† (57)	0.43‡ (126)
Abdominal fat (%)	0.36‡ (183)	0.31* (57)	0.36‡ (126)	0.40‡ (183)	0.31† (57)	0.43‡ (126)
IAAT (cm^2)	0.41‡ (181)	0.38† (55)	0.39‡ (126)	0.46‡ (181)	0.37† (55)	0.51‡ (126)
DXA trunk:total ratio	0.34‡ (183)	0.32* (57)	0.32‡ (126)	0.36‡ (183)	0.39† (57)	0.39‡ (126)

NOTE. Sample sizes are given in parentheses.
*$P \leq .05$; †$P \leq .01$; ‡$P \leq .001$.

Five ANCOVAs were performed to investigate the effect of ethnicity on CETP activity in univariate analyses (Table 6). In model 1 only ethnicity was entered into the model, while in the other 4 models ethnicity was entered as a covariate plus the following variables: model 2—age and height, as these variables differed significantly between Hispanic and Anglo women; model 3—triglyceride concentration and total cholesterol:HDL cholesterol ratio, as both were significant predictors of CETP activity in the multiple regression analysis; model 4—each measure of body fat distribution to examine the effect of regional body composition; and model 5—each measure of total body fat to examine the effect of total body composition. Models 4 and 5, including the variables intra-abdominal adipose tissue and total body fat (kg), respectively, are presented as these variables had the highest correlations with CETP activity. As expected, ethnicity was a significant variable in the model of CETP activity (model 1). Ethnicity remained significant after age and height were added as covariates in model 2. Once significant predictor variables (triglyceride concentration and total cholesterol:HDL cholesterol ratio) were introduced in model 3, ethnicity was no longer a significant variable in the model. Additional analyses revealed that the addition of any regional body composition measure or lipoprotein variable resulted in ethnicity no longer being a significant variable in the model of CETP activity (model 4 as an example). However, the inclusion of any total body composition measure resulted in the persistence of ethnicity as a significant variable in the model of CETP activity (model 5 as an example).

DISCUSSION

There is very little information reported in the literature regarding LCAT and CETP activities in postmenopausal women, or ethnic influences on their activities. Study of factors that may affect CETP and LCAT, and thus HDL cholesterol metabolism and reverse cholesterol transport, would be helpful when designing interventions to reduce the risk of coronary heart disease in this population. This study was designed to

Table 5. Stepwise Multiple Regression Including Age, Height, Total and Regional Body Composition, and Lipids and Lipoproteins With CETP and LCAT Activities as Dependent Variables

Significant Variables	Regression Coefficient	Standardized Beta	Adjusted R^2	SEE (μg/mL · h)
Dependent variable: CETP				
Ethnicity	4.38†	0.29		
Triglyceride concentration	0.02*	0.26		
Total:HDL cholesterol ratio	2.51†	0.36		
Intercept	−9.63†		0.46	6.42
Dependent variable: LCAT				
Ethnicity	3.83†	0.30		
Total:HDL cholesterol ratio	2.40‡	0.41		
BMI	−0.41*	−0.26		
WHR	32.22‡	0.38		
Intercept	−7.44		0.45	5.42

NOTE. Variables included in analysis: ethnicity, HRT, total cholesterol, HDL cholesterol, LDL cholesterol, total cholesterol:HDL cholesterol, triglycerides, LDL oxidation, age, height, weight, BMI, total body fat (% and kg), IAAT, intra-abdominal fat (% and kg), WHR, skinfold trunk:total ratio, and DXA trunk:total ratio (add waist).
*$P \leq .05$; †$P \leq .01$; ‡$P \leq .001$.

Table 6. ANCOVA Examining Ethnic Differences in CETP Activity

Model Variables	Ethnicity *P* Value	Model *P* Value
Model 1		
Ethnicity	.02	.02
Model 2		
Age		
Height	.03	.07
Model 3		
Triglyceride concentration		
Total:HDL cholesterol ratio	.30	.001
Model 4		
IAAT	.39	.001
Model 5		
Total body fat (kg)	.03	.001

NOTE. Ethnicity was included in all models. Significance level is reported in "Ethnicity *P* Value" column. All variables listed were significant at $P < .05$.

examine plasma CETP and LCAT activities in postmenopausal Hispanic and Anglo women, as well as plasma and body composition factors that may influence their activities. Hispanic women had (1) higher mean plasma CETP activity, (2) lower plasma HDL cholesterol concentrations, (3) higher total cholesterol:HDL cholesterol ratios, (4) higher plasma triglyceride concentrations, and (5) greater susceptibility of LDL particles to oxidation compared to Anglo women. Additionally, Hispanic women had a significantly greater relative deposition of body fat on the trunk and intra-abdominally than did Anglo women, even after adjusting for total body fat, as has been reported previously.[12,13] Positive associations between intra-abdominal adipose tissue and plasma CETP and LCAT activities were found in both Hispanic and Anglo women.

In the present study, plasma CETP activity was consistently related to plasma lipids and lipoproteins, as has been found previously in the literature.[35] Correlations were much higher between CETP activity and triglyceride concentrations than for other lipoproteins. This might be expected as very–low-density lipoprotein (VLDL)-triglycerides have been found to be a rate-limiting substrate for CETP activity in normolipidemic subjects.[36,37] CETP activity has been found previously to be negatively related to HDL cholesterol and HDL cholesterol:total cholesterol ratio in humans.[38] These results are consistent with those of the present study, as HDL cholesterol was negatively associated with CETP activity and positively related to total cholesterol:HDL cholesterol ratio (an inverse of the previously mentioned ratio). CETP concentrations have also been found to be positively associated with LDL cholesterol concentrations, which is also consistent with the present study results.[39,40]

The significant difference in plasma CETP activity between Anglo and Hispanic women was eliminated after accounting for intra-abdominal adipose tissue and other regional measures of body fatness, but not with measures of total body fatness suggesting a role of abdominal fat in regulating CETP activity. Adjusting for measures of body fat distribution has also been found previously to eliminate ethnic differences in serum lipids and lipoproteins between Mexican American and Anglo women, 25 to 64 years of age.[12] The positive relationship between CETP activity and the abdominal distribution of body fat seen in this study might be expected as CETP is synthesized in adipose tissue.[41] Arai et al reported a significantly higher CETP activity and mass and a lower HDL cholesterol concentration in obese men and premenopausal women compared to normal-weight subjects.[42] These researchers also reported a significant positive correlation between CETP activity and BMI, body fat ratio, and subcutaneous fat area by computed tomography scan imaging, and a negative correlation with visceral:subcutaneous fat ratio.[42] Weight reduction resulted in a reduction in both plasma CETP protein and activity. The authors, however, suggested that subcutaneous fat, not visceral fat, may play a role in regulating CETP levels.[42]

Ethnic differences were not found with plasma LCAT activity. LCAT activity was positively related to intra-abdominal adipose tissue; however, the mechanism by which adipose tissue may play a role in LCAT regulation is unclear, as LCAT is synthesized in the liver and located on HDL particles. Significant positive correlations were found with plasma LCAT activity and total cholesterol, total cholesterol:HDL cholesterol ratio, LDL cholesterol, the oxidative susceptibility of LDL particles, and triglycerides. The best predictors of LCAT activity were ethnicity, total cholesterol:HDL cholesterol ratio, BMI, and WHR. In a previous study of 79 men, plasma LCAT concentrations were positively related to plasma triglyceride and LDL cholesterol concentrations.[43] These authors found LCAT concentrations to be negatively related to HDL_2 concentrations and positively related to HDL_3 concentrations resulting in nonsignificant correlation with HDL cholesterol levels, which may also be relevant to the present study as we report a small but significant negative relationship with HDL cholesterol.

The function of LCAT is to esterify cholesterol on HDL particles, resulting in incorporation of additional tissue cholesterol onto the particles. The resulting cholesterol ester is then transferred to apoprotein B–containing particles. A higher concentration of cholesterol esters in the HDL particle would be expected to limit LCAT activity. The correlation between LCAT activity and triglyceride level was of a higher magnitude than correlations between LCAT activity and other plasma variables in the present study. Significant correlations between LCAT activity and triglyceride levels may be related to the basic mechanism by which reverse cholesterol transport is thought to function. As mentioned earlier, and increase in triglyceride levels is associated with an increase in CETP activity,[36,37] which would subsequently result in a reduction in cholesterol ester on the HDL lipoprotein as it is exchanged for VLDL-triglyceride. A reduction in particle cholesterol ester would, thus, provide a higher ratio of LCAT substrate to product, resulting in increased LCAT activity. The positive relationship between LCAT activity and triglyceride levels may also be the result of a decrease in lipoprotein lipase activity; however, this enzyme was not investigated in the present study.

Limited data are available regarding the oxidative susceptibility of LDL particles in Mexican Americans.[44] In the present study an increased oxidative susceptibility of LDL particles was found in Hispanic women compared to Anglo women, suggesting that Hispanic women are at increased coronary heart disease risk due to the potential modifications of LDL particles. We are unable to make conclusions as to the origin of the ethnic

difference in the susceptibility of LDL particle to oxidation in this group of women. The results may be due to dietary differences or may be genetic in nature.

A significant positive relationship was found between CETP activity and LDL particle oxidation susceptibility. The oxidative susceptibility of LDL particles has previously been found to be negatively related to LDL particle size with small, dense LDL particles being more susceptible to oxidation than large particles.[45] As an increased CETP activity may result in smaller LDL particles,[46] oxidative susceptibility of LDL particles would be expected to be positively related to CETP activity. Alternatively, an increase in CETP activity is expected to enrich LDL particles with triglycerides relative to cholesterol ester. The triglyceride particles are susceptible to hydrolysis by lipases, which would result in a reduction in LDL particle size. The function of CETP in both LDL particle size and susceptibility of LDL particles to oxidation are subjects for further investigation.

Plasma LCAT activity was also positively related to LDL oxidation in the present study. The results from in vitro studies suggested that LDL oxidation inhibits LCAT activity in a dose-dependent manner.[47,48] The results of the present in vivo study differ from the results of the above in vitro studies. The roles that CETP and LCAT may play in the oxidative susceptibility of LDL particles are beyond the scope of this report and require further investigation.

Age, postmenopausal years, and years of HRT use were not significantly related to CETP or LCAT activities. Plasma LCAT activity in postmenopausal women has not been previously reported in the literature. Sacks et al reported no effect of estrogen on plasma CETP concentration in a small group of postmenopausal women, although there was a significant increase in HDL cholesterol concentration with estrogen use.[49] HRT use in postmenopausal women in the present study had no effect on either CETP or LCAT activities. This would suggest that other factors, such as body composition, are important in regulating HDL metabolism in postmenopausal women.

In summary, CETP and LCAT activities were consistently related to plasma lipids and lipoproteins and total and regional body composition. CETP activity was positively related to the intra-abdominal deposition of body fat, possibly due to CETP synthesis by adipose tissue. In this study, Hispanic postmenopausal women appear to be at higher risk for coronary heart disease as suggested by lower plasma HDL cholesterol, higher triglyceride concentrations, increased oxidative susceptibility of LDL particles, and a greater regional deposition of adipose tissue. These particular factors may also contribute to higher CETP activity in postmenopausal Hispanic women.

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